EFFECT OF SPLENECTOMY AND BLOCKING THE KUPFFER CELLS WITH INK ON MITOTIC ACTIVITY OF HEPATOCYTES AFTER HEPATECTOMY

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UDC 612.35.014.3:612.6-06:612.412

Splenectomy was performed on one group of rats two days before partial hepatectomy. A second group of rats received an injection of 2 ml 1% India ink into the spleen daily for four days before partial hepatectomy in order to block the reticuloendothelial system of the liver. Increased mitotic activity of the hepatocytes was observed in both experimental groups later than in the control.

KEY WORDS: Partial hepatectomy; splenectomy; ink block; mitotic activity.

The classical investigations of B. Fischer, V. G. Garshina, and F. M. Lazarenko showed that proliferative growth of the epithelium depends on the state of the underlying mesenchymal cells [3, 5, 6]. The question of interaction between epithelium and connective tissue assumes even greater importance in connection with the clearly demonstrated role of tissue induction in organ formation during the period of embryogenesis [4]. Investigations of regeneration and epithelization of the wound surface have revealed delayed restoration of the epithelial component when activity of the underlying connective-tissue cells is inhibited [7]. Some workers speak of the stimulant effect of lymphoid tissue on mitotic activity of the hepatocytes [1, 2]. It has been shown, for instance, that after injection of a suspension of spleen cells from hepatectomized animals into intact mice the mitotic index of the hepatocytes and, in particular, of the reticuloendothelial cells of the liver rises considerably. These observations raise the question of interaction between Kupffer cells and hepatocytes and the possible stimulant effect of lymphoid tissue on the mitotic activity of the hepatocyte of the regenerating liver through the Kupffer-cell apparatus.

To examine these problems the mitotic activity of the hepatocytes was studied after partial hepatectomy in rats with the reticuloendothelial system of the liver blocked and after splenectomy.

EXPERIMENTAL METHOD

Experiments were carried out on 105 noninbred male albino rats weighing 120-140 g. Splenectomy was performed two days before partial hepatectomy under ether anesthesia through a lateral incision. The Kupffer cells were blocked by injecting 2 ml% India ink once a day for four days into the animals through a lateral incision into the spleen under ether anesthesia. This method was developed so as to avoid diffuse spread of the ink throughout the body when injected into rats via the caudal vein. Partial hepatectomy was performed two days later. A group of animals receiving 2 ml physiological saline by injection into the spleen daily for four days, followed two days later by hepatectomy, and animals undergoing partial hepatectomy alone, were used as the control. Considering that the results of both controls were very similar, these groups of animals will subsequently be considered together. Partial hepatectomy on all groups of animals was carried out through a midline incision, and two lobes of the liver (60-70%) were removed. The animals were killed 24, 36, 48, 72, and 96 h and eight days after the operation at the same time of day (9:00 -9:30 a.m.); 5 rats were killed at each time in the experimental series, and 6-8 rats in the control. Car-

Department of Pathological Anatomy, I. P. Pavlov First Leningrad Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR A. P. Avtsyn.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 80, No. 7, pp. 91-93, July, 1975. Original article submitted September 20, 1974.

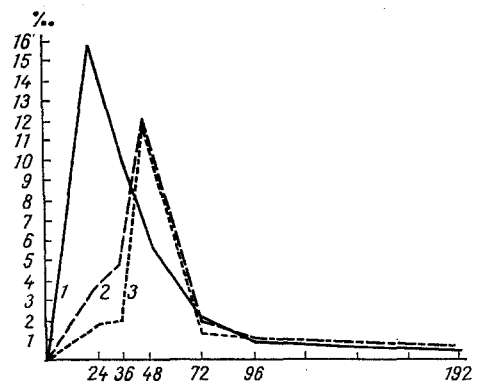

Fig. 1. Mitotic activity of hepatocytes at various times after partial hepatectomy: 1) partial hepatectomy; 2) hepatectomy after splenectomy; 3) hepatectomy after blocking Kupffer cells. Abscissa, time after operation (in h); ordinate, mitotic activity (in $^0/_{00}$).

noy's fluid was used for fixation. Mitoses were counted in 8000 hepatocytes in each animal in sections 5 μ in thickness, stained with hematoxylin–eosin.

EXPERIMENTAL RESULTS

Mitotic activity in the control group was highest 24 h after partial hepatectomy (mean 15.9% of mitoses), and it fell gradually until the 8th day ($0.3^0/_{00}$). When partial hepatectomy was performed after splenectomy, marked inhibition of mitotic activity of the hepatocytes was observed in the early stages after removal of part of the liver: $3.7^0/_{00}$ after 24 h and $5^0/_{00}$ after 36 h. The maximal increase was observed after 48 h ($11.9^0/_{00}$), with a further decrease until the 8th day (Fig. 1). The curve of mitotic activity in the second experimental group (partial hepatectomy with blocking of the reticuloendothelial tem of the liver with ink) resembles that when partial hepatectomy followed splenectomy, but in this case the degree of inhibition of mitotic activity was greater in the early stages ($1.9^0/_{00}$ of mitoses after 24 h and $2.1^0/_{00}$ of mitoses after 36 h). Later, as in the splenectomized animals, an increase in the number of mitoses was observed until 48 h ($12.0^0/_{00}$) and a gradual decrease until the 8th day (Fig. 1).

Inhibition of mitotic activity of the hepatocytes was thus observed in the early stages after partial hepatectomy following splenectomy and blocking of the reticuloendothelial cells of the liver, with an increase in mitotic activity later (11.9 and $12.0^0/_{00}$ of mitoses respectively by 48 h compared with $15.7^0/_{00}$ in the control). Inhibition of mitotic activity in this period can evidently be explained by adaptation, during which the functions of the Kupffer-cell apparatus and lymphoid tissue are restored. Considering the similarity between the curves of mitotic activity in animals undergoing partial hepatectomy after splenectomy and after blocking of the reticuloendothelial cells of the liver, it can be postulated that the action of the lymphoid tissue is effected through the Kupffer cells. The possibility of disturbance of diffusion of metabolic products and biologically active substances through the hepatic sinus, the walls of which were loaded with ink particles, cannot be ruled out. However, whatever the mechanism of inhibition of mitotic activity in the hepatocytes, the fact of close interconnection between the epithelial and connective-tissue components and also between the epithelium and lymphoid system of the body as a whole is manifested sufficiently completely in the liver regenerating after hepatectomy.

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